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Effects of Two Triazole Seed Treatments, Triticonazole and Triadimenol, on Growth and Development of Wheat

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Abstract: The triazole fungicides, triadimenol and triticonazole, applied as seed treatments at various rates on spring wheat, affected plant growth, shoot development, and root axis production. The main growth regulatory effects were reductions in lengths of the coleoptile, the first leaf and the subcrown internode. A marked effect on tiller appearance occurred with high rates of triadimenol. Modification of root-system development included a reduction in the number of seminal roots, increased outgrowth of roots associated with the coleoptilar node, and a reduction of roots at the first foliar node (correlated with reduced production of Tiller 1). Seed treatment effects on both shoot and root development illustrated the close relationship between these plant structures. Both fungicides induced both beneficial and deleterious effects on wheat growth and development, but the magnitude of deleterious effects was less with triticonazole than with triadimenol.

Key words: triazole fungicides, triadimefon, triticonazole, seed treatment, wheat.

1 INTRODUCTION

Triazole derivatives are used as fungicides and plant growth regulators in agriculture and as antimycotic and antibacterial agents in medicine. They inhibit the conversion of lanosterol to ergosterol or other 4,14-demethyl sterols in higher fungi (sterol biosynthesis inhibitors : SBI), interfering with membrane function.[1–3]

Because these chemicals are systemically translocated in plants and have a broad spectrum of fungitoxicity, the triazole derivatives are widely used in agriculture, by foliar or seed treatment application. Triadimenol was the first triazole fungicide to be used as a translocated seed treatment.[4,5] The fungicide is effective against major cereal diseases caused by seed-borne pathogens, powdery mildew, rusts, and eyespot.[6–8] Recently another triazole derivative, triticonazole, has been found to possess high activity against many cereal diseases.[9,10]

Many plant growth regulating properties have also been described for SBI fungicides.[3,11] Fletcher and Hofstra[12] indicated that plant regulatory effects of triadimefon are more pronounced after application on seeds or young seedlings than on mature plants. Phytotoxic effects of triadimenol and its precursor, triadimefon, include delayed emergence, reduced surface area of coleoptiles and leaves, growth retardation, reduced root length, and tillering aberrations.[6,7,13–16]

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Another SBI, imazalil, reduced the length of the subcrown internode of wheat and increased the number of coleoptile-node tillers.[17] Effects of triadimenol seed treatment on development of root axes have not been described, and effects of triticonazole on plant growth and development have not yet been evaluated.

The main purpose of this study was to investigate the effect of triticonazole and triadimenol seed treatments on growth and development of wheat (*Triticum aesticum* L.) shoots and roots.

2 MATERIALS AND METHODS

2.1 Plant growth and seed treatments

The experiment was performed at the Columbia Basin Agricultural Research Center, 13 km NE of Pendleton, Oregon in Pacific Northwest Agronomic Zone 2.[18] The Center is in a 430-mm precipitation zone with a Walla Walla silt loam soil.

Seeds of the soft white spring wheat cultivar Wakans were treated with either triadimenol 150 g litre^{-1} SC (Baytan 15 Flo, Bayer AG) or triticonazole 200 g litre^{-1} SC (RP 400727; 'Premis,' Rhône-Poulenc). Each fungicide was applied at doses of 0·075, 0·15, 0·3, 0·6, 1·2, and 2·4 g AI kg^{-1} seed. Triticonazole was also tested at 0·0375 and 4·8 g kg^{-1} seed. All fungicide treatments were applied in 24 ml of aqueous suspension per kg of seeds. In current practice, the standard for triadimenol seed treatment is 0·3 g AI kg^{-1} seed. The application rate for triticonazole in intensive agriculture is 1·2 g AI kg^{-1} seed.[19] For control treatments, an equivalent amount of water was applied to the seed.

Seeds were planted (50 kg ha^{-1}, *c.* 72 seeds m^{-1} of row) within one week after treatment, on 25 April 1991. Seed depth was about 5 cm. The experiment was a randomized complete block design with four replications for each of 15 treatments. Plots measured 1·5 × 5 m and contained five rows of plants spaced at 30-cm intervals.

2.2 Data collected

Four assessments were made of plant growth and development. Shoot development was assessed as described by Masle-Meynard and Sebillotte[20] and Klepper *et al.*,[21] with emphasis on tiller sequence of appearance. Root system development was monitored as described by Klepper *et al.*[22] (Fig. 1). This system, which assigns numbers to root axes according to the node from which they arise, describes the direction of root growth with a letter (A, B, X, Y). This system permitted determination of effects of the chemicals on production of individual seminal roots (−2 and −1 nodes), coleoptilar node (0 node) roots, and crown (1, 2, 3 nodes) roots.

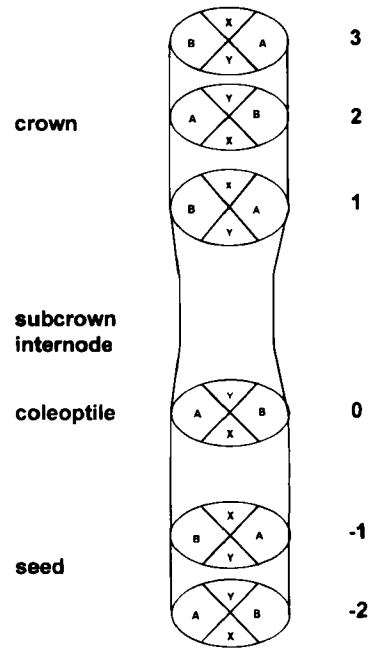

Fig. 1. Origins of four potential root axes per node at six successive nodes of a wheat stem. A different root originates in each of four quadrants at each node. The roots designated A and B develop together, followed by the roots designated X and Y. Roots from nodes −1 and −2 are 'seminal' roots and the presence of the scutellum prevents the outgrowth of axes in the −2X and −1Y quadrant, giving the usual total of six seminal axes. Roots from the coleoptile node (0) are located at planting depth and are often confused with seminal roots. Roots from nodes 1, 2 and 3 are considered 'coronal' or crown roots.

2.2.1 *First assessment*

Numbers of emerged seedlings were counted 13 days after planting (8 May) at Haun stage (main stem leaf number) of 1·3 in the control.[23] The measurement was taken on plants in four 20-cm lengths of row in each plot. Twenty plants from the three center rows were collected to assess seedling growth and development, including measurements of coleoptile length, plant height from the soil line to the tip of the longest leaf, width of leaf one and seminal root development (presence of radical, −2A, −2B, −1A, −1B, −1X and −2Y axes). Haun stage was also assessed and dry weights of aerial and underground parts were determined.

2.2.2 *Second assessment*

A second sampling was made 26 days after planting (21 May) when the control was at Haun stage 3.1. Data collection included number of seminal roots, presence of roots associated with the coleoptilar node, i.e. 0A, 0B,

0X and 0Y, and presence of roots 1A and 1B at the first foliar node. Other measurements included plant growth stage, presence of Tiller One (T1) at the first foliar node, length of subcrown internode, plant height, and dry weights of aerial and underground parts.

2.2.3 Third assessment

A third assessment was made 60 days after planting (24 June) when the control was at Haun stage 9·6. Data collection included development of individual tillers (T0 to T5), Haun stages of main stem and each successive tiller, plant height and dry weight, and numbers of seminal and coronal roots.

2.2.4 Fourth assessment

On 22 July, the number of heads was counted in two 1-m lengths of row.

3 RESULTS

3.1 Emergence

The number of emerged plants 13 days after sowing (Table 1) was significantly reduced when seeds were treated at the highest rate of triadimenol (2·4 g kg^{-1} seed). All other treatments of triadimenol and triticonazole (data not presented) did not significantly effect seedling emergence.

TABLE 1
Effects of Triadimenol on Emergence of Wheat Plants 13 Days after Planting

Dose (g AI kg^{-1} seed)	*Number of emerged plants per meter of row*[a]
0	55·3bc
0·075	55·6bc
0·15	61·3bc
0·3	51·6bc
0·6	49·1bc
1·2	41·3ab
2·4	33·4a

[a] Means followed by the same letter do not differ significantly at $P < 0.05$ by Newman Keuls.

3.2 Growth parameters

Plant height 13 days after planting (Table 2) was reduced by all rates of triadimenol and at rates of triticonazole greater than 0·15 g kg^{-1} seed. As plants aged, growth suppression occurred only at very high rates of triadimenol and there was no growth suppression for triticonazole. In fact, plant height was increased

TABLE 2
Effects of Triticonazole and Triadimenol Seed Treatments on Wheat Plant Growth[a]

Treatment and dose (g AI kg^{-1} seed)	*Length of coleoptile (mm) 13 days after planting*	*Plant height (cm) Days after planting*			*Width of first leaf (mm) 13 days after planting*	*Length of subcrown internode (mm) Days after planting*	
		13	*26*	*60*		*26*	*60*
Untreated check	36ab	6·6a	13·5b	47·6bcd	3·2d	12·0a	10·3a
Triticonazole (0·0375)	33bc	6·3ab	14·6a	47·6bcd	3·4cd	6·0c	5·5b
Triticonazole (0·075)	37a	6·3ab	14·7a	49·1abc	3·4cd	7·8b	4·7b
Triticonazole (0·15)	32cd	6·2abc	15·1a	50·4a	3·4cd	2·3de	2·3c
Triticonazole (0·3)	31cde	5·6cde	15·1a	48·8abc	3·4cd	2·4de	2·4c
Triticonazole (0·6)	34bc	5·5de	14·7a	48·6abc	3·4cd	2·4de	1·6c
Triticonazole (1·2)	32cd	6·0bcd	15·1a	49·6a	3·5bc	2·1de	1·8c
Triticonazole (2·4)	34bc	5·9bcd	14·9a	49·4ab	3·5bc	1·9de	1·3c
Triticonazole (4·8)	33bc	5·6cde	14·2a	49·8a	3·4c	3·1d	0·8c
Triadimenol (0·075)	38a	5·9bcd	15·1a	49·6a	3·4cd	1·1de	0·6c
Triadimenol (0·15)	30def	5·7bcde	15·1a	49·7a	3·6abc	0·1e	0·0c
Triadimenol (0·3)	32cd	5·2e	14·9a	48·4abc	3·7ab	0·0e	0·0c
Triadimenol (0·6)	28ef	4·7f	14·5a	47·3cd	3·7ab	0·0e	0·0c
Triadimenol (1·2)	29ef	4·4g	12·8c	46·1d	3·8a	0·0e	0·0c
Triadimenol (2·4)	27f	3·7g	11·8d	46·0d	3·7ad	0·0e	0·0c

[a] Means within columns followed by the same letter do not differ significantly at $P < 0.05$ by Newman Keuls.

by triticonazole and the low rates of triadimenol at the second assessment. Later there were no differences. Both chemicals restricted the length of the first leaf but leaf width was increased by the three highest rates of triticonazole and by all the lowest rate of triadimenol. Coleoptile length was reduced by all except the lowest rate of triadimenol and by several intermediate rates of triticonazole.

A large decrease in length of subcrown internode occurred with both chemicals at the second and third assessments (Table 2). Triadimenol essentially eliminated elongation of the subcrown internode, and plants grown from the triticonazole-treated seeds had significantly shorter subcrown internodes than those from non-treated seeds. Hence, the crown was deeper after seed treatments by both fungicides.

The data for dry weights are not presented. A slight decrease of shoot dry weights was correlated with the delay of emergence observed after seed-treatment with high rates of triadimenol at the first assessment. Later, there were no differences in dry weights of plants grown from treated or non-treated seeds.

3.3 Shoot development

The pattern of appearance of tillers was modified in plants from triadimenol-treated seeds (Table 3). Most of the T1s and, in some cases, T2s (tillers at the base of the first, L1, and the second, L2, leaves, respectively) were skipped when high rates of triadimenol were applied. No significant tiller loss was observed with triticonazole (data not presented).

Inhibition of production of the first two tillers was compensated by a higher production of T4 and T5. In the control treatment, plants did not form any T5s, whereas 45% of plants grown from seeds treated with the highest rate of triadimenol had a T5. This compensation produced similar numbers of heads in treated or non-treated plots when counted 22 days before harvest (data not presented).

TABLE 3
Effects of Triadimenol Seed Treatment on Tiller Formation in Wheat 60 Days after Planting

Dose (*g AI kg*$^{-1}$ *seed*)	*Plants with tillers (%)*[a]				
	T1	*T2*	*T3*	*T4*	*T5*
0	85a	97a	98a	62ab	0b
0·075	82a	100a	93a	53ab	3b
0·15	78a	100a	97a	63ab	2b
0·3	58abc	100a	100a	53ab	2b
0·6	60abc	98a	97a	83ab	7b
1·2	40bc	85b	98a	93a	7b
2·4	35c	85b	98a	93a	45a

[a] Means within columns followed by the same letter do not differ significantly at $P < 0{\cdot}05$ by Newman Keuls.

There was no effect of seed treatments on Haun stages of the main stem, and most tillers (Table 4). The highest rate of triadimenol caused accelerated development of T3 and T4 and caused T4 to have one leaf more than in the control. Because of the lack of T5 in the control, it was difficult to demonstrate any advance of this tiller, but T5 of plants grown from seeds treated with the highest rate of triadimenol had 2.2 leaves, comparable to the T4 of plants grown from non-treated seeds.

3.4 Root system development

3.4.1 Seminal roots

At the first assessment, the radicle and −2A and −2B axes (roots associated with the scutellar node) were

TABLE 4
Effects of Triadimenol Seed Treatment on Leaf Number (Haun stage) of Main Stem and Successive Tillers of Wheat 60 Days after Planting[a]

Dose (*g AI kg*$^{-1}$ *seed*)	*Main stem*	*T1*	*T2*	*T3*	*T4*
0	9·6ab	6·1ab	5·6ab	4·2b	2·5bc
0·075	9·4bc	6·3ab	5·6ab	4·1b	2·4bc
0·15	9·6ab	6·5a	6·0a	4·4ab	2·4bc
0·3	9·3bc	6·0ab	5·6ab	4·2b	2·6bc
0·6	9·5abc	6·1ab	5·4b	4·4ab	2·7bc
1·2	9·5abc	5·9ab	5·6ab	4·5ab	3·0b
2·4	9·7a	5·5b	5·5ab	4·7a	3·5a

[a] Means within columns followed by the same letter do not differ significantly at $P < 0{\cdot}05$ by Newman Keuls.

TABLE 5
Effects of Triticonazole and Triadimenol Seed Treatments on Seminal Root Development in Wheat[a]

Treatment and dose (g AI kg^{-1} seed)	*Percentage of plants with −1X root (13 days after planting)*	*Total number of seminal roots 26 and 60 days after planting*	
		26	*60*
Untreated check	44a	5·3a	5·5a
Triticonazole (0·0375)	36ab	5·1abc	5·2ab
Triticonazole (0·075)	23ab	5·2ab	5·2ab
Triticonazole (0·15)	28ab	5·0abc	5·1bc
Triticonazole (0·3)	21ab	5·1abc	5·0bcd
Triticonazole (0·6)	20ab	5·1abc	5·1bc
Triticonazole (1·2)	20ab	5·0abc	5·1bc
Triticonazole (2·4)	26ab	5·1abc	5·0bcd
Triticonazole (4·8)	31ab	5·2ab	5·2ab
Triadimenol (0·075)	25ab	5·0abc	5·0bcd
Triadimenol (0·15)	26ab	4·9bc	5·0bcd
Triadimenol (0·3)	19ab	4·8c	4·7d
Triadimenol (0·6)	20ab	4·9bc	4·8cd
Triadimenol (1·2)	17b	5·0abc	4·9bcd
Triadimenol (2·4)	23ab	5·0abc	5·0bcd

[a] Means within columns followed by the same letter do not differ significantly at $P < 0.05$ by Newman Keuls.

always present (data not presented). Roots associated with the epiblast-node (−1A and −1B) were produced in 65% to 90% of the plants and were not affected by seed treatments. In contrast, differences were noted for the −1X axis (Table 5) which is the sixth axis to appear in wheat.[24] This root was presented on 44% of the seedlings grown from untreated seed, in as few as 20% (not statistically different) in triticonazole treatments and

TABLE 6
Effects of Triticonazole and Triadimenol Seed Treatments on Development of Coleoptilar Node Roots and Crown Roots in Wheat 26 Days after Planting[a]

Treatment and dose (g AI kg^{-1} seed)	*Total number of roots associated with nodes 0, 1, 2, 3, 4 ...*	*Mean number of roots at node 0*	*Mean number of roots at node 1*
Untreated check	1·8bc	1·1def	0·7a
Triticonazole (0·0375)	2·1ab	1·5abc	0·6abc
Triticonazole (0·075)	2·0abc	1·3cde	0·7a
Triticonazole (0·15)	2·4a	1·9a	0·6abc
Triticonazole (0·3)	2·1ab	1·7abc	0·4bcdef
Triticonazole (0·6)	2·1ab	1·7abc	0·4abcde
Triticonazole (1·2)	2·3a	1·7abc	0·6ab
Triticonazole (2·4)	2·0abc	1·7abc	0·3cdefg
Triticonazole (4·8)	2·0abc	1·5abc	0·5abcd
Triadimenol (0·075)	1·7bc	1·5abc	0·2efg
Triadimenol (0·15)	1·8bc	1·4abcd	0·3cdefg
Triadimenol (0·3)	1·8bc	1·4abcd	0·3defg
Triadimenol (0·6)	1·5c	1·4abcd	0·1fg
Triadimenol (1·2)	1·1d	1·1def	0·1g
Triadimenol (2·4)	1·0d	0·9f	0·1g

[a] Means within columns followed by the same letter do not differ significantly at $P < 0.05$ by Newman Keuls.

17% in one of the triadimenol treatments. Total numbers of seminal roots did not differ among treatments 13 days after planting (data not presented). Evaluations of seminal roots 26 and 60 days after planting corroborated the earlier assessment that −1X and −2Y axes were often missing (Table 5).

3.4.2 Coleoptilar node roots

Roots associated with the coleoptilar node (node 0) for 26-day-old plants were mainly 0A and 0B. Results indicated that triticonazole treatment at nearly all rates or the lowest rates of triadimenol enhanced the production of coleoptilar node roots (Table 6).

3.4.3 Crown roots

At Haun stage 3.1 (26 days after planting), half of the plants grown from non-treated seeds had crown roots (roots 1A and 1B associated with node 1). The total number of roots at this assessment was significantly higher in the control than with any triadimenol seed treatment (Table 6).

Although some significant effects of seed treatments on total number of roots (seminal roots excepted) were measured 26 days after planting (Table 6), differences among treatments did not occur 60 days after planting, at which time the total number of roots varied from 27 to 33.

4 DISCUSSION

Actual rate of fungicide application to each seed varies considerably during commercial seed treatment procedures. Some of the rates examined in this study were extremely high, e.g. almost 10 times the standard rate of triadimenol. Under these conditions, the two seed treatments affected many aspects of plant growth and development. Some negative effects on early tillering were compensated with later tillers and, then, no effect on numbers of heads is found. Some authors have even reported that the reduction in tiller formation may increase heads per meter of row.[25,26] In this study, high rates of triadimenol delayed emergence. Even when tested at the standard rates, this chemical has occasionally been reported[6] to decrease coleoptile elongation and, hence, emergence. The reduction in height of plants appeared to be due mostly to regulatory effects at the beginning of growth (coleoptile, first leaf). Reduction in surface area of primary leaves has also been reported for beans drenched with triadimefon[14] and after cereal seed treatments with triadimefon or triadimenol.[13] In contrast, triticonazole and low concentrations of triadimenol induced a slight increase of height of 26-day-old plants. Stimulation of shoot growth has also been described after seed treatment of wheat with very low rates of triadimefon.[25]

Both fungicides caused a drastic shortening of the subcrown internode and, hence, a marked effect on crown depth. Chinn *et al.*[17] obtained similar results with wheat plants grown from imazalil-treated seeds. This shortening of the subcrown internode might reduce emergence and stand establishment.

Triadimenol, but not triticonazole, had a marked effect on tiller development. With high rates of triadimenol, about 50% of the plants produced T1 and 90% produced T2. Tillering perturbations caused by seed treatments have been reported.[16,25] Compensation of later tillering for reduction in early tillering, as shown by a higher production of T4 and T5 in this study, has been consistently noticed in field experiments.[7,15] Although the risk of abortion of a tiller is reported to be higher with the youngest tillers,[27] our results relating to numbers of heads showed that in this experiment, most T4s and T5s produced a head. In addition to the impact of triadimenol seed treatment on production of tillers, this fungicide also modified growth stages of tillers with delay of the oldest and advance of the youngest. In our study, reduced percentages of T1 and T2 could have been associated with the absence of a subcrown internode in triadimenol-treated plants. Without a subcrown internode, crown depths on seedlings from treated seed would have been greater and hence would have simulated a situation of deep seeding. Deep seeding reduces or delays emergence of T0 and T1.[20,26]

This study revealed that modifications of root system development were induced by seed treatment. Effects of triadimefon on growth of bean[28] and bluegrass[29] have been described. On cereals, the only observations of such effects are related to the length of roots and do not describe which axes are affected.[13] Rickman *et al.*[24] reported that cultivar and seed size were the main factors that affected the number of seminal roots. The present experiment indicates that fungicide seed treatments also affect seminal root development. The last seminal roots developed more rarely on plants grown from treated seeds and, as a result, the total number of seminal roots produced was lower.

Root development at the 0-node is quite irregular, and is affected by plant and environmental factors.[24,30] Seed treatments appeared in the present study to be a factor which favorably influences the outgrowth of roots associated with the coleoptilar node. Although such results have not previously been reported for triazoles, they have been reported for the imidazole imazalil which, when applied as seed treatment, induced an increase in the number of the coleoptilar node tiller roots and tillers.[17] The trend in the present study was similar although the number of coleoptilar node tillers was not enhanced. It is possible that the materials and energy saved by the failure to elongate the subcrown

internode could have become available for the production of these roots at the coleoptilar node.

Both tiller production and root development at node 1 were reduced by triadimenol seed treatment. This correlation is supported by results of Klepper *et al.*,[22] who reported that the 'X' root, the third root at a node, was present only if the tiller associated with this node was produced. Root axis numbers 60 days after planting did not differ among treatments, suggesting that the compensation in number of tillers was also associated with a compensation in root number.

The present study provides information about the physiological effects of two triazole fungicides applied as seed treatments. It demonstrates that these two chemicals had both beneficial and detrimental effects on wheat growth and development. Negative impacts of seed treatments were less for triticonazole than for triadimenol. The system described by Klepper *et al.*[22] allowed us to quantify the close relationships between shoot and root development in wheat and to reveal how chemicals applied to seeds can disturb this balance. This study could also help create a better understanding of whether morphological changes in addition to fungitoxic effects could directly affect the protection of the plant from certain diseases.

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